REMARKS

Claims 1-19 were pending in the subject application. Applicant has hereinabove canceled claims 4, 5 and 15-19 and amended claims 1-3 and 6-14. Accordingly, upon entry of this Amendment claims 1-3 and 6-14 will be pending and under examination.

Applicant maintains that these amendments to the claims do not raise any issue of new matter, and that these claims are supported by the specification as originally filed.

Objection to the Specification

In the May 31, 2006 Office Action the Examiner objected to the specification since for the proper designation for the identifying a sequence number, i.e. "SEQ ID NO:", was not used to identify the sequences disclosed in the subject application.

Further in the May 31, 2006 Office Action, the Examiner objected to the specification since it contains numerous typographical errors.

In response, applicant first notes that the typographical errors contained in the paragraph on page 14 were previously amended on April 21, 2004 by the applicant. In addition, applicant has hereinabove amended the specification to correct typographical errors and to correctly identify all sequence numbers disclosed in the subject application with the sequence identifier "SEQ ID NO:". Applicant maintains that these amendments to the specification do not raise any issue of new matter, and that these amendments are supported by the specification as originally filed. Accordingly, applicant respectfully requests the Examiner to remove these objections to the specification.

Objection to the Claims

In the May 31, 2006 Office Action the Examiner objected to the claims since claim 3 contained a typographical error, claims 6-10 were drawn to non-elected matter and claims 11 and 14 did not contain the proper designation for the identifying a sequence number, i.e. "SEQ ID NO:", claim 8 was missing a space after the word "claim" and claims 6-14 were objected to as allegedly being in improper form.

In response, applicant has hereinabove amended the claims to correct the typographical errors, correctly identify all sequence numbers disclosed in the subject application with the sequence identifier "SEQ ID NO:" and place the claims in an acceptable form. Applicant maintains that these amendments to the claims do not raise any issue of new matter, and that these amendments are supported by the specification and claims as originally filed. Accordingly, applicant respectfully requests the Examiner to remove these objections to the claims.

AUGUST 29, 2006 Interview

Applicant wishes to thank Examiner Monshipouri and Examiner Rooke for their time and consideration during their August 29, 2006 telephonic interview with the undersigned and Dr. Peng Liang. During the interview, Examiner Monshipouri indicated an certain obviousness concerns of the current invention over Mohler et al. which is disclosed on page 3 of the subject application. Examiner Monshipouri alleged that, from the disclosure of Mohler and the existence of commercially available dimeric proteins, it would be obvious for one skilled in the art to produce a trimeric protein. In addition, Examiner Monshipouri wished to see experimental evidence that the trimeric sTNFR

created by the current invention exhibits surprisingly and unexpected increased binding over dimeric sTNFR.

Applicant addresses Examiner Monshipouri's comments in his response to the May 31, 2006 Office Action obviousness rejection below.

Rejection Under 35 U.S.C.§102(b)

In the May 31, 2006 Office Action, the Examiner rejected claims 1-3 under 35 U.S.C. §102 as allegedly anticipated by Vuorio et al. (PCT International Publication No. WO 97/17988). The Examiner alleged that Vuorio et al. disclose a fusion collagen using a method comprising a culturing host cells transformed with a recombinant DNA expression vector, and recovering the fusion protein from the cell culture.

In response to the Examiner's rejection, applicant respectfully traverses on the ground that the cited reference does not recited all the elements of the pending claims.

Briefly, the pending claims recite a method for generating a secreted soluble trimeric fusion protein, comprising a) creating a DNA construct comprising a transcriptional promoter linked to a template encoding a signal peptide sequence followed by in-frame fusion to a non-collagenous polypeptide to be trimerized, which in turn is joined in-frame to a mammalian polypeptide capable of self-trimerization which is heterologous from the non-collagenous polypeptide to be trimerized; (b) introducing said DNA construct into a eukaryotic cell; (c) growing said host cell in an appropriate growth medium under physiological conditions to allow the secretion of a trimeric fusion-protein

encoded by said DNA sequence; (d) isolating said trimeric fusion protein from the culture medium of said host cell.

Vuorio et al. only discloses fusion proteins containing different collagens. Vuorio et al. does not disclose a polypeptide capable of self-trimerization, such as collagen, fused to noncollagenous polypeptides.

In addition, the fusion polypeptides disclosed in Vuorio et al. would form part of an insoluble collagen matrix and would not be soluble as the polypeptides of the pending claims. As noted on page 5 of the specification as filed, procollagen is flanked by a non-collagenous domain called the C-propeptide. This domain is processed proteolytically upon secretion of the procollagen, which triggers the mature collagen to form an insoluble matrix. Accordingly, Vuorio et al. does not disclose all the elements of the pending claims.

In view of the applicant's amendments to the claims, applicant respectfully requests the Examiner to remove this ground of rejection.

Rejection Under 35 U.S.C.§103(a)

In the May 31, 2006 Office Action, the Examiner rejected claims 1-3 under 35 U.S.C. §103(a) as allegedly obvious in light of Gruber et al. (U.S. Patent No. 6,617,431). The Examiner indicated that Gruber et al. teaches the use of a recombinant nucleotide sequence containing a cDNA coding for one or several mammalian collagen chains and elements enabling a plant cell to produce the collagen chains. The Examiner alleged that based upon the disclosure of Gruber et al. it would have been obvious to one skilled in the art at the time the invention was made to

design a method to generate a secreted trimeric fusion protein, such as collagen, because a method of creating a DNA comprising a sequence of choice, and introducing that DNA construct into a host cell, and growing a host cell in a medium, and isolating a protein of interest, is disclosed by Gruber et al. The Examiner further alleged that one skilled in the art would be motivated to design the instant method of generating different collagen fusion proteins because different protein combinations, such as trimeric collage, could be created by that method.

In response to the Examiner's rejection, applicant respectfully traverses on the ground that a prima facie case of obviousness has not been established.

Under MPEP §2143, to establish a prima facie case of obviousness the cited reference must teach or suggest every element of the claim.

Gruber et al. does not teach or suggest all the elements of the pending claims. Gruber et al. does not teach or suggest a method of producing a fusion of a polypeptide capable of self-trimerization, such as collagen, fused to noncollagenous polypeptides. Gruber only discloses the production of one or several mammalian collagen chains in a plant cell. In addition, the fusion polypeptides disclosed in Gruber et al. would form part of an insoluble collagen matrix and would not be soluble as the polypeptides of the pending claims. As noted on page 5 of the specification as filed, procollagen is flanked by a non-collagenous domain called the C-propeptide. This domain is processed proteolytically upon secretion of the procollagen, which triggers the mature collagen to form an <u>insoluble</u> matrix.

Therefore, Gruber et al. does not teach or suggest all the elements of the pending claims. In view of the applicant's amendments to the claims, applicant respectfully requests the Examiner to remove this ground of rejection.

August 29, 2006 Interview Obviousness Concern

As noted above, during an August 29, 2006 telephonic interview, Examiner Monshipouri indicated an obviousness concern of the current invention over Mohler et al. which is disclosed on page 3 of the subject application. Mohler et al. discloses an IgG-Fc fusion for protein dimerization that is used to make commercially available dimeric TNFRII (e.g. Enbrel® from Amgen). Examiner Monshipouri alleged that, from the disclosure of Mohler and the existence of commercially available dimeric proteins, it would be obvious for one skilled in the art to produce a trimeric protein. The Examiner appeared to base her obviousness concern on an understanding that Mohler et al. teaches that the more multivalent a soluble receptor is, the more potent it can be in binding to its ligand. Therefore, since one skilled in the art would be motivated to produce a stronger binding soluble receptor, one skilled in the art would be motivated to produce a trimeric soluble receptor (three-binding sites) over a dimeric soluble receptor, and would further be motivated to produce a tetrameric soluble receptor (four binding site).

In addition, Examiner Monshipouri wished to see experimental evidence that the trimeric sTNFR created by the current invention exhibits surprisingly and unexpected increased binding over dimeric sTNFR.

In response, without conceding that Mohler et al. discloses a prima facie basis of obviousness, applicant respectfully addresses the Examiner's concern by pointing out that the subject invention is not obvious in light of Mohler et al.

As noted on page 3 of the subject application, Mohler et al. discloses one of the current strategies for treating diseases caused by the overactivation of cell receptors. Receptor "decoys" consisting of soluble extracellular ligand-binding domains of cell receptors are used to intercept and bind to excess ligand to prevent the overactivation of cell receptors. Thus, receptor decoys are designed with increased binding affinities to ligands to increase the interception and binding of excess ligand.

This rationale of designing stronger receptor decoys is found in Mohler et al. on page 1549 as follows: "Given predominantly trimeric nature of TNF and the apparently requirement for cross-linking of cell-surface TNFR for signal transduction, it is likely that dimeric soluble receptor constructs should possess a higher affinity for TNF and therefore function as a considerably more potent competitive inhibitors than monomeric sTNFR."

Mohler et al. lead to the commercially successful dimeric sTNFRs such as Enbrel®, a soluble dimeric decoy consisting of a single TNF-α receptor fused to an immunoglobin (IgG). Despite the success of Enbrel®, the ligand to which it binds, TNF-α, exists as a fusion of three identical polypeptides. Each of these identical subunits can bind to the TNF-α receptor. Thus, high amounts of Enbrel® must be injected into a patient to bind enough TNF-α ligand to have a therapeutic effect.

Given the rationale suggested in Mohler et al. to seek more potent inhibitors and the limitations of Enbrel® stated above, the non-obviousness of the current invention is best illustrated by the fact that 13 years after the creation of dimeric sTNFR such as Enbrel®, no one has come up with a way for creating secreted soluble TNFR (or any other secreted protein, for that matter) in trimeric form.

Other protein trimerization domains, such as GCN4 from yeast (Yang, X. et al, 2000), fibritin from bacteria phage T4 (Frank, S. et al., 2001) and aspartate transcarbamoylase from *Escherichia coli* (Chen, B. et al., 2004), have been described previously in the art. Copies of these references are attached to the accompanying Information Disclosure Statement for the Examiner's review. These previously described systems allow the trimerization of heterologous proteins, but none of these trimerizing proteins are mammalian in nature, nor are they naturally secreted proteins. As such, any trimeric fusion proteins would have to be made intracellularly, which not only may fold incorrectly for naturally secreted proteins such as soluble receptors, but also makes purification of such fusion proteins from thousands of other intracellular proteins difficult. Moreover, the fatal drawback of using such non-mammalian protein trimerization domains (e.g. from yeast, bacteria phage and bacteria) for trimeric biologic drug design will be their immunogenicity (antibody generating) in mammals such as humans, rendering such fusion proteins ineffective within weeks after injecting into the subject. Thus, the prior art, alone or in combination, does not disclose a method for producing a suitable trimeric fusion protein as claimed in the subject application.

Examiner Monshipouri also based her obviousness concern on the understanding that Mohler et al. teaches or suggests that the more multivalent a soluble receptor is, the more potent it can be in binding to its ligand. In other words, the Examiner alleged that it would be obvious to one skilled in the art to add as many ligand binding sites as possible to soluble receptor since a greater number of binding sites would produce soluble receptors with greater binding affinities.

The applicant, and those skilled in the art, would disagree with this generalization of Mohler et al. For example, a tetrameric sTNFR, which would be tetravalent (four binding sites) cannot theoretically have a higher affinity to its trivalent ligand TNF (3 binding sites) than a trimeric sTNFR (3 binding sites). From a geometric point of view, a structure of 2-fold symmetry (such as a dimeric or tetrameric sTNFR) theoretically cannot perfectly overlay with a trimeric structure (such as TNF), which has 3-fold symmetry (See Fig. 1 for illustration). From protein structure point of view, a trimeric sTNF should have the highest binding affinity to its trimeric ligand, TNF. Therefore, it is not obvious to those skilled in the art that generating soluble receptors with more binding sites produces stronger binding affinities.

As for the Examiner's desire to see experimental evidence that the trimeric sTNFR created by the current invention exhibits surprisingly and unexpected increased binding over dimeric sTNFR, applicant notes that there is no requirement that the claimed invention must be better than or provide specific advantages over what was known in the art. This is not the test of obviousness.

Although the applicant respectfully maintains that such evidence is not relevant for the validity of the current invention, which offers a new platform for trimerization of any secreted protein, applicant is pleased to address the Examiner's concern without conceding the correctness thereof.

Applicant points to the examiner to Figures 4A and 4B which show the affinity of the trimeric TNFR (sTNF-RII-T2) disclosed in the subject application to TNF in the standard TNF bioassay. As seen on page 25, the trimeric TNFR binding affinity to TNF in the standard TNF bioassay was 10-100 times more potent in TNF binding than that of the published binding affinity of dimeric sTNFR.

In due respect, applicant truly thanks the examiners for the critical analysis of our invention, and hope with this revision and clarification, the examiners could now appreciate the validity of the current invention.

INFORMATION DISCLOSURE STATEMENT

In accordance with his duty of disclosure under 37 C.F.R. §1.56 applicant directs the Examiner's attention to the following references which are listed on the attached Form PTO/SB/08B (**Exhibit A**) and attached hereto as **Exhibits 1-3.**

Applicant files this Information Disclosure Statement under 37 C.F.R. §1.97 (c)(2), before the mailing of a final office action on the merits accompanied by the fee of ONE HUNDRED EIGHTY DOLLARS ($180.00) set forth in 37 C.F.R. §1.17 (p). Accordingly, applicant requests that this Information Disclosure Statement be considered.

1. Yang, X. et al., "Characterization of Stable, Soluble Trimers Containing Complete Ectodomains of Human Immunodeficiency Virus Type I Envelope Glycoproteins," J. Virol. 74:5716-5725 (2000) (**Exhibit 1**);

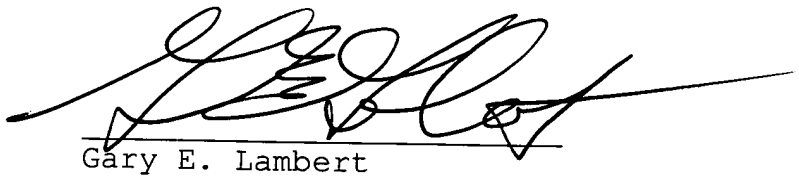
2. Frank, S. et al., "Stabilization of Short Collagen-like Triple Helices by Protein Engineering," J. Mol. Biol. 308:1081-1089 (2001) (**Exhibit 2**); and

3. Chen, B. et al., "A Chimeric Protein of Simian Immunodeficiency Virus Envelope Glycoprotein gp140 and *Escherichia coli* Aspartate Transcarbamoylase," J Virol. 78:4508-4516 (2004) (**Exhibit 3**).

If a telephone interview would be of assistance in advancing prosecution of the subject application, applicant's undersigned attorney invite the Examiner to contact them at the number provided below.

No fee, other than the $180.00 for filing an Information Disclosure Statement enclosed by check, is deemed necessary in connection with the filing of this Amendment and Information Disclosure Statement. If any additional fee is required, authorization is hereby given to charge the amount of any such fee to Deposit Account 12-0115.

Respectfully submitted,



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